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IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of: Stanley et al.

Application No. 10/029,430

Filed: December 18, 2001

For: ANTIGEN PREPARATION AND USE

Examiner: To be assigned

Date: March 28, 2002

Art Unit: 1648

CERTIFICATE OF MAILING

I hereby certify that this paper and the documents referred to as being attached or enclosed herewith are being deposited with the United States Postal Service on March 28, 2002 as First Class Mail in an envelope addressed to: COMMISSIONER FOR PATENTS, BOX MISSING PARTS/SEQUENCE, P O BOX 2327, Arlington, Virginia, 22202.

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RESPONSE TO NOTICE TO FILE CORRECTED APPLICATION PAPERS AND SECOND PRELIMINARY AMENDMENT

This is in response to the Notice to File Corrected Application Papers dated January 28, 2002, for which a two-month period for reply was set, making a response due on or before March 28, 2002.

Prior to examination of the above-referenced application, please amend the application as follows:

In the specification:

Please replace page 7 as originally filed with the following:

--(N)-terminus of full-length L1 was fused to amino acid residues 50-65 from E1 protein of HPV16, and (C) the amino (N)-terminus of full-length L1 was fused to amino acid residues

50-65 from E1 protein HPV16 and additionally, amino acid residues 384-403 from E1 protein of HPV16 were fused at the N-terminus corresponding to sequence (B).

For generation of recombinant (A), the sequence of HPV16 L1 ORF was amplified by polymerase chain reaction (PCR) from a total DNA extract of Wl 2, an HPVl6 episome-containing cell line (Stanley M, Brown HM, Appleby M, and Minson AC (1989) "Properties of a non-tumorigenic human keratinocyte cell line", Int J Cancer 43, 672-676) using a forward primer in which a BgI 11 restriction sequence was included (underlined) 5'-GCT GCA AGA TCT ATG TCT CTT TGG CTG CCT AG-3' **(SEQ ID NO: 1)**.

For generation of recombinant (B), a DNA nucleotide sequence encoding the El 50-65 sequence plus a BgI 11 restriction sequence (underlined) was introduced just in front of the L1 coding sequence as a forward primer, as follows: 5'-GCT GCA AGA TCT ATG GTA GAT TTT ATA GTA AAT GAT AAT GAT TAT TTA ACA CAG GCA GAA TCT CTT TGG CTG CCT AGT GAG-3' **(SEQ ID NO: 2)**.

For generation of recombinant (C), a nucleotide sequence-encoding El amino acids 384-403 was introduced just in front of El 50-65 coding sequence of the last-mentioned construct, using a forward primer with a flanking BgI 11 restrictional sequence (underlined): 5'-GCT GCA AGA TCT ATG TAC GAT AAT GAC ATA GTA GAC GAT AGT GAA ATT GCA TAT AAA TAT GCA CAA TTG GCA GAC GTA GAT TTT ATA GTA AAT GAT-3' **(SEQ ID NO: 3)**.

These forward primers were paired with the same reverse primer in which a Not 1 restriction site was included (underline). Reverse: 5'-GAT CTA GCG GCC GC TTA CAG CTT ACG CTT CTT GCG TTT-3' **(SEQ ID NO: 4)**.

Following 30 cycles of amplification, the DNA products (e.g. of about 1.7kb in size) were gel purified, GENECLEAN (TM) excised, digested with restriction enzymes of BgI 11 and Not 1 and sub-cloned into baculovirus transfer vector pBacAK8 (Clontech) which had

been pre-digested with BamHI and Not I restriction enzymes. The recombinant plasmids for cases (A), (B) and (C) were examined by sequencing in per-se known manner (Pharmacia (TM) kit).

Generation of recombinant baculoviruses:

Insect cells of Spodoptera frugiperda (sf2l) were grown in 30 mm dishes until 80% confluent, at 27°C with TNMFH medium (Sigma) supplemented with 1 0% foetal--

In the Abstract:

Please add the following abstract as page 15 of the specification:

--ABSTRACT

Modified virus-like particles (VLPs) can comprise fusion proteins having sequence from a major coat protein of papovavirus, e.g. L1 protein and HPV 16 or 18, in which the N-terminal of the sequence derived from the major coat protein is fused to a further peptide sequence. The VLPs can contain a full sequence of an L1 protein, or an L1 sequence with an N-terminal deletion, or an L1 sequence with an amino acid substitution mutation, and optionally a C-terminal L1 sequence deletion. The peptide sequence fused to the N-terminal can be immunogenic, e.g. from a protein of a pathogen such as a virus. The further peptide sequence can provide a binding domain for affinity purification of the VLP. Modified VLPs can retain the native conformation of the VLP structure while also presenting to the immune system of a subject immunized with the modified VLPs an epitope present on an N-terminal extension of the major coat protein sequence. Corresponding polynucleotides, expression vectors, plasmids, vectors and cells containing such polynucleotides are disclosed.--

REMARKS

This Response and Second Preliminary Amendment is submitted to recite sequence numbers for the sequences set forth in the application and to correct a typographical error, and to add the Abstract. No new matter is added. Entry of this amendment is respectfully requested.

CONCLUSION

If any minor matters remain to be discussed prior to examination, the Examiner is invited to contact the undersigned at the telephone number listed below.

Respectfully submitted,

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Marked-up Version of Amended Specification
Pursuant to 37 C.F.R. §§ 1.121(b)-(c)

In the specification:

Please amend page 7 of the specification as shown on the attached marked-up version of page 7, which is attached hereto. Please note that the nucleic acid sequences underlined on lines 9-10, 12, 19 and 23 were underlined on page 7 as submitted, and are not new additions.

In the abstract:

Please add the attached Abstract as page 15 of the specification.

(N)-terminus of full-length L1 was fused to [aminoacid] amino acid residues 50-65 from E1 protein of HPV16, and (C) the amino (N)-terminus of full-length L1 was fused to amino acid residues 50-65 from E1 protein HPV16 and additionally, [aminoacid] amino acid residues 384-403 from E1 protein of HPV16 were fused at the N-terminus corresponding to sequence (B).

For generation of recombinant (A), the sequence of HPV16 L1 ORF was amplified by polymerase chain reaction (PCR) from a total DNA extract of W1 2, an HPV16 episome-containing cell line (Stanley M, Brown HM, Appleby M, and Minson AC (1989) "Properties of a non-tumorigenic human keratinocyte cell line", Int J Cancer 43, 672-676) using a forward primer in which a Bgl 11 restriction sequence was included (underlined) 5'-GCT GCA AGA TCT ATG TCT CTT TGG CTG CCT AG-3' **(SEQ ID NO: 1)**.

For generation of recombinant (B), a DNA nucleotide sequence encoding the El 50-65 sequence plus a Bgl 11 restriction sequence (underlined) was introduced just in front of the L1 coding sequence as a forward primer, as follows: 5'-GCT GCA AGA TCT ATG GTA GAT TTT ATA GTA AAT GAT AAT GAT TAT TTA ACA CAG GCA GAA TCT CTT TGG CTG CCT AGT GAG-3' **(SEQ ID NO: 2)**.

For generation of recombinant (C), a nucleotide sequence-encoding El amino acids 384-403 was introduced just in front of El 50-65 coding sequence of the last-mentioned construct, using a forward primer with a flanking Bgl 11 restrictional sequence (underlined): 5'-GCT GCA AGA TCT ATG TAC GAT AAT GAC ATA GTA GAC GAT AGT GAA ATT GCA TAT AAA TAT GCA CAA TTG GCA GAC GTA GAT TTT ATA GTA AAT GAT-3' **(SEQ ID NO: 3)**.

These forward primers were paired with the same reverse primer in which a Not 1 restriction site was included (underline). Reverse: 5'-GAT CTA GCG GCC GC TTA CAG CTT ACG CTT CTT GCG TTT-3' **(SEQ ID NO: 4)**.

Following 30 cycles of amplification, the DNA products (e.g. of about 1.7kb in size) were gel purified, GENECLEAN (TM) excised, digested with restriction enzymes of Bgl 11 and Not 1 and sub-cloned into baculovirus transfer vector pBacAK8 (Clontech) which had been pre-digested with BamHI and Not I restriction enzymes. The recombinant plasmids for cases (A), (B) and (C) were examined by sequencing in per-se known manner (Pharmacia (TM) kit).

Generation of recombinant baculoviruses:

Insect cells of Spodoptera frugiperda (sf2l) were grown in 30 mm dishes until 80% confluent, at 27°C with TNMFH medium (Sigma) supplemented with 10% foetal